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SYNTHESIS AND BIOLOGICAL ACTIVITY OF SOME 1-HYDROXY-3-AMINOALKYLADAMANTANES AND THEIR DERIVATIVES

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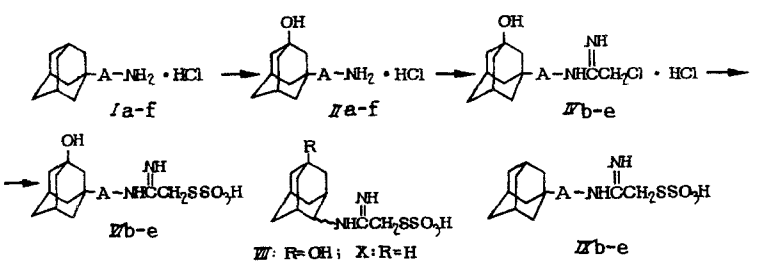
Some adamantanes have been found to possess a wide spectrum of biological activity [2, 4, 7, 8], drugs such as remantadine and adapromine having already found application for the prophylaxis and early treatment of influenza. There have been reports of the radioprotectant effects of N-adamantyl derivatives of 2-acetamidinethiosulfuric and thiophosphoric acids [9, 12].

In order to assess their biological activity, and to establish structure-activity relationships, we have now obtained some adamantanes containing a hydroxy group in the ring, such compounds having different hydrophobic properties, thereby potentially modifying the uptake and distribution of the compounds in the body.



Throughout A: a) bond, b) CH_2O, c) $(CH_2)_2$ d) $(CH_2)_3$ e) $CH(CH_3)$, f) $CH(CH_2CH_3)$

The 3- and 5-hydroxy-derivatives of 1- and 2-aminoadamantane and their derivatives have previously been obtained by treatment with a mixture of nitric and sulfuric acids [6]. It was shown that hydroxylation also occurred smoothly and cleanly when the amino-group in the 1-position was separated from the ring by a hydrocarbon chain A, the aminoalkyladamantane hydrochlorides (Ia-f) affording 71-85% yields of the hydroxylated compounds 3-amino-, 3-aminomethyl-, 3-aminoethyl-, 3-aminopropyl-, 3-(1-aminoethyl)-, and 3-(1-aminopropyl)-1-hydroxyadamantanes (IIa-f). The structures of (IIb-f) were confirmed by PMR spectroscopy. Similarly, 2-aminoadamantane hydrochloride gave 1-hydroxy-4-aminoadamantane (III). Treatment of the amines (IIb-e) with chloroacetonitrile in methanol in the presence of catalytic amounts of sodium methoxide afforded the chloracetamidines (IVb-e). Under the same condi-

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TABLE 1. Data for Compounds Obtained

Compound	Yield, %	mp, °C	Empirical formula
IIa	90	>340[a]	$C_{10}H_{17}NO \cdot HCl$
IIb	76	110—112	$C_{11}H_{19}NO \cdot 0{,}25H_2O$
HC IIb		274—276	$C_{11}H_{19}NO \cdot HCl$
IIc	73,5	73—75	$C_{12}H_{21}NO$
HC IIc		292—295	$C_{12}H_{21}NO \cdot HCl \cdot 1{,}5H_2O$
IId	71	109—111	$C_{13}H_{23}NO$
HC IId		179—183	$C_{13}H_{23}NO \cdot HCl \cdot 0{,}25H_2O$
IIe	78,8	111—123	$C_{12}H_{21}NO$
HC IIe		301—303	$C_{12}H_{21}NO \cdot HCl$
HC IIa	85	282—284	$C_{13}H_{23}NO \cdot HCl$
III	78	258—265	$C_{10}H_{17}NO$
HC III		>340[b]	$C_{10}H_{17}NO \cdot HCl$
IVb	85	259—260 decomp	$C_{13}H_{21}ClN_2O \cdot HCl \cdot 0{,}25H_2O$
IVc	78	220—222	$C_{14}H_{23}ClN_2 \cdot HCl$
IVd	84		$C_{15}H_{25}ClN_2O \cdot HCl \cdot 0{,}75H_2O$
IVe	84	249—250 decomp	$C_{14}H_{23}ClN_2O \cdot HCl$
V	92,5	250—251 decomp	$C_{12}H_{19}ClN_2O \cdot HCl$
VIII	83,5	245—248	$C_{14}H_{23}ClN_2 \cdot HCl$
VIb	92,5	206—208 decomp	$C_{13}H_{22}N_2S_2O_4$
VIc	58	172—178	$C_{14}H_{24}N_2S_2O_4$
VId	63	166—170	$C_{15}H_{26}N_2S_2O_4 \cdot 2H_2O$
VIe	88	220—225	$C_{14}H_{24}N_2S_2O_4 \cdot 2H_2O$
VII	92	210—220 decomp	$C_{12}H_{20}N_2S_2O_4 \cdot 0{,}5H_2O$
IXe	74	192—196	$C_{14}H_{24}N_2S_2O_2 \cdot 0{,}5H_2O$

[a]Lit. mp 356-358°C [11]

[b]Lit. mp 350°C [10]

TABLE 2. Cyctotoxic and Antiviral Activity of Test Compounds

Compound	MTC, μg/ml		ED_{50} in CAM, μg/ml			
	CEF	CAM	A/Bankok/1/79	CTI	B/Hong Kong/72	CTI
Ib	31,25	250	62,5	4,0	62,5	4,0
Ic	7,8	62,5	15,6	4,0	15,6	4,0
Id	3,9	31,2	7,8	4,0	16,6	2,0
IIa	125	300	150	2,0	150	2,0
IIb	62,5	300	200	1,5	200	1,5
IIc	62,5	300	150	2,0	150	2,0
IId	62,5	300	200	1,5	200	1,5
IIe	125	400	100	4,0	100	4,0
IIf	62,5	300	75	4,0	150	2,0
III	62,5	300	200	1,5	200	1,5
VIb	62,5	250	75	3,3	100	2,5
VIc	62,5	125	50	2,5	50	2,5
VIe	62,5	250	25	10	150	1,6
IXe	7,8	7,8	2	3,9	4	1,9
Remantadine (Ie)	75	200	5	40	*	
Adapromine (If)	65	150	5	30	10	15

*Remantadine in a concentration of 1/2 MTC failed to inhibit the multiplication of influenza virus B/Hong Kong/72 in CAM culture.

tions, the amine (III) was converted into N-(1-hydroxy-4-adamantyl)-2-chloroacetamidine (V). A similar derivative could not be obtained from 1-hydroxy-3-aminoadamantane (Ia) either by treatment of the amine hydrochloride with acetonitrile, or by reaction of the free base (Ia) with ethyl or methyl 2-chloracetimidate. Reaction of the amidines (IVb-e) and (V) with sodium thiosulfate in aqueous methanol gave the thiosulfuric acid derivatives (VIb-e) and (VII).

Similarly, from (Ie) there was obtained the chloroacetamidine (VIII), which was then converted into S- N-[1-(1-adamantyl)ethyl]acetamidino thiosulfuric acid (IXe).

The antiviral and radioprotectant activity of the compounds was examined.

EXPERIMENTAL (CHEMISTRY)

The progress of the reactions was followed and the purity of the products checked by TLC on Silufol plates in the system ethyl acetate—ethanol—hexane—ammonia (3:3:1:2), developer iodine vapor.

IR spectra were obtained on a Perkin-Elmer 398 instrument, and PMR spectra on a Bruker WM-250 spectrometer with DSS as internal standard. The data for the products are given in Table 1. The elemental analyses agreed with the calculated values.

1-Hydroxy-3-aminoalkyladamantanes (IIa-f). To 100 ml of conc. sulfuric acid was added 10 ml of 60% HNO_3, and 0.05 mole of the 1-aminoalkyladamantane hydrochloride (Ia-f) was then added portionwise at 10-15°C. The mixture was stirred for 6-20 h, poured on to ice, basified, and extracted with chloroform. The extract containing the 1-hydroxy-3-aminoalkyladamantane base was dried, the solvent removed, and the residue dissolved in alcohol and treated with alcoholic HCl. The (II) hydrochlorides were isolated either by evaporation of the alcoholic solutions, or by precipitation with ether. The IR spectral data (KBr disks) showed the presence in the compounds (IIa-f) of OH and NH_3^+ groups (absorption at 2900-3500 cm^{-1}), and new bands at 1045-1080 cm^{-1} (C=O), absent in the original compounds (Ia-f).

PMR spectral data (δ, ppm): (IIb), 1.58 t (1H, H^2), 1.48-1.52 m (6H, H^4, H^6, H^{10}), 2.26 m (2H, H^5, H^7), 1.60-1.78 m (4H, H^8, H^9), 2.77 s (2H, CH_2); (IIc), 1.54 m (2H, H^2), 1.41-1.48 (6H, H^4, H^6, H^{10}), 2.17 m (2H, H^5, H^7), 1.58-1.72 m (4H, H^8, H^9), 1.44-1.48 m (2H, $AdCH_2$), 2.87-2.97 m (2H, CH_2NH_2); (IId), 1.54 br (2H, H^2), 1.38-1.46 m (6H, H^4, H^6, H^{10}), 2.17 br (2H, H^5, H^7), 1.58.173 m(4H, H^8, H^9), 1.14-1.24 m (2H, $AdCH_2$), 1.55-1.7 m (2H, CH_2), 2.95+ (2H, CH_2NH_2) (J 7.63 Hz); (IIe), 1.57 m (2H, H^2), 1.51 m (6H, H^4, H^6, H^{10}), 2.27 m (2H, H^5, H^7), 1.61-1.77 m (4H, H^8, H^9), 1.24 d (3H, CH_3) (J 7.02 Hz), 3.09 q (1H, CH) (J 7.02 Hz); (IIf), 1.58 br (2H, H^2), 1.51-1.53 m (6H, H^4, H^6, H^{10}), 2.27 br (2H, H^5, H^7), 1.61-1.77 m (4H, H^8, H^9), 1.01+ (3H, CH_3) (J 7.33 Hz), 1.90 d.t (2H, CH_2) (J 7.33 Hz, 3.05 Hz), 2.84 d (1H, CH) (J 3.05 Hz).

N-(1-Hydroxyadamant-3-ylethyl)-2-chloroacetamide Hydrochloride (IVc). To a solution of NaOMe (from 0.02 g of Na and 40 ml of methanol) was added 1.6 g (0.02 mole) of chloracetonitrile. The mixture was kept for 30 min, 2.03 g (0.01 mole) of (IIc) hydrochloride added, stirred for 45 min, concentrated to half its volume, 200 ml of ether added, and the solid filtered off and washed with ether to give 2.4 g of the hydrochloride (IVc). Compounds (IVb-e) and (V) were obtained similarly.

S-[N-(1-Hydroxyadamant-3-ylmethyl)acetamido]thiosulfuric Acid (VIb). A solution of 2.94 g (0.01 mole) of the hydrochloride (IVb) and 2.73 g (0.01 mole) of $Na_2S_2O_3 \cdot 5H_2O$ in a mixture of 30 ml of methanol and 10 ml of water was boiled under reflux for 1.5 h. The mixture was then evaporated to dryness with the addition of two portions of absolute ethanol. The residue was dissolved in DMF, and kept overnight in the refrigerator. The sodium chloride which separated was filtered off, and the filtrate treated with an excess of ether. The solid which separated was filtered off and washed with ether to give 3.1 g of (VIb). Compounds (VIc-e), (VII), and (IXe) were obtained similarly.

EXPERIMENTAL (BIOLOGY)

The antiviral activity of the adamantanes was examined against influenza viruses A/Bangkok/1/79 (H3N2) and B/Hong Kong/72 and Sindbis avian virus. Toxicities were determined in a primary trypsinized culture of chick embryo fibroblasts (CEF) and a surviving culture of chick embryo chorioallantoic membrane (CAM). The data obtained were use to establish the maximum tolerated concentrations (MTC) of the compounds [3]. The samples were dissolved immediately before the tests at an initial concentration of 1000 μg/ml, from which were prepared twofold dilutions. The antiviral activity was assessed relative to influenza viruses A and B in CAM culture by the reduction in the infective titer and the hemagglutinin titer, and relative to the Sindbis virus in CAM culture by the inhibition of platelet formation. Activities were expressed as the ED_{50} values, defined as the minimum effective concentration (dose) inhibiting by 50% the multiplication of 100 infective doses of the test virus [5]. The antiviral activity of the test compounds was compared with those of remantadine and adapromine. The results are shown in Table 2.

TABLE 3. Radioprotectant Activity of Test Compounds

Compound	Mode of administration (15 min before irradiation	LD_{50}, mg/kg	Radioprotectant activity	
			dose, mg/kg	% survival
Id*	ip	79	28	10
IIf*	ip	>1000	123	0
VI b	ip	150	50	0
IXb**	ip	25	10	93
VIc	ip	100	30	0
IXc**	ip	40	10	27
VId	ip	200	70	0
IX**	ip	8	2,5	40
VIe	ip	120	40	0
VII	ip	200	70	0
VII	ip	200	70	0
	po	1200	400	6,6
X**	ip	38	10	100
	po	280	200	0
IXe	ip	20	10	26,5

*Compound tested by V. I. Kulinskii at the Krasnoyar Institute of Medicine.

**Literature results [11]

Note. ip indicates intraperitoneal, and po peroral.

The radioprotectant activity of the compounds was examined in mice, strain C57BL/6. Aqueous solutions of the compounds in a volume of 0.2 ml were administered to the animals 15 min before irradiation. The mice were irradiated in an IGUR-1 apparatus at a dose of 800 cGy and a dose rate of 1.2 cGy/sec, $LD_{96-100/30}$. The toxicities of the compounds were determined in mongrel white mice weighing 22-25 g. LD_{50} values were calculated as in [1]. The test results are shown in Table 3.

As will be seen from Table 2, all the test compounds inhibited influenza virus A/Bangkok/1/79 in concentrations of 1/2-1/4 of the MTC. The highest activity against this virus was shown by (VIe), the ED_{50} value for which was ten times less than the maximum tolerated concentration. Also noteworthy is (IIe), the ED_{50} value of which against influenza viruses A and B was 100 μg/ml [chemotherapeutic index (CTI) 4.0]. Multiplication of virus A/Bangkok/1/79 was inhibited to the same extent (CTI 4.0) by (Ib-d), (IIe, f), and (IXd), and of these (Ib) and (Ic) inhibited multiplication of the influenza B virus to the same extent.

None of the test compounds showed activity against the Sindbis avian virus.

Although none of these adamantane derivatives showed antiviral activity approaching that of remantadine, a well known antiinfluenza drug (Ie, CTI 40), the presence of a hydroxy group in the adamantane nucleus reduced toxicity and extended the antiviral spectrum of activity, thus encouraging further research in this area.

It will be seen from Table 3 that introduction of a hydroxyl group into the adamantane nucleus results in the almost total loss of radioprotectant activity. The nonhydroxylated analogs of the test compounds are included in this Table for comparison.

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